Post-exercise provision of 40 g of protein during whole body resistance training further augments strength adaptations in elderly males

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Abstract

Background: In elderly populations, low intake of dietary protein exacerbates the effects of sarcopenia and anabolic resistance, so, protein supplementation to maximise muscle protein synthesis, has the potential to be an effective intervention strategy. **Aim:** This study aimed to determine the effects of a low and high dose of protein, ingested immediately post-exercise, during a resistance training programme in novice elderly males. **Method:** Twenty-four healthy elderly (70.5±5.1, years) Caucasian males were recruited for this study (body mass: 92.4±14.9 kg; fat free mass: 61.4±7.6 kg; fat mass: 31.2±10.2 kg). After exclusion criteria were applied, 18 males participated. Participants continued with their normal habitual dietary intake and were allocated into two matched groups, which were then randomly assigned to either a low (20 g) or high (40 g) dose intervention. Following the determination of 1 repetition maximums (1RM) participants completed 10 x $3d^{-1}$ wk resistance training and consumed protein supplements immediately following exercise. **Results:** Significantly greater improvements in chest press ($p = 0.014$, ηp^2 0.34) shoulder press ($p = 0.005$, ηp^2 0.43) and leg extension strength ($p = 0.014$, ηp^2 0.34), were observed following the 40 g dose, resulting in larger performance improvements of 19.1, 21.1, and 16.1% respectively, compared to the 20 g dose. **Conclusion:** Findings suggest that ingesting 40 g of protein following resistance exercise, produces greater responses to training than 20 g, in novice elderly males, and may be an important nutritional strategy which should be used when prescribing resistance exercise in the elderly.

Keywords: Muscle protein synthesis, exercise prescription, anabolic resistance, sarcopenia, muscle strength, fat free mass.

Introduction

Improved living conditions, and treatment for infectious and cardiovascular diseases, have improved longevity, resulting in a greater number of older and elderly people in developed societies (Suzman et al., 2015). Despite this increase, there has been little effect on active life expectancy (Keeler et al., 2010, Katz et al., 1983), an important predictor of quality of life, which is not indicative of genuine developments in longevity (Noale et al., 2012). A major public health issue (Gomes et al., 2017) in older populations is sarcopenia which has been shown to be associated with debilitating loss of skeletal muscle function (Koopman, 2010).

In sedentary individuals >70 years old, muscle function declines of 60% have been observed compared with their younger counterparts (Close et al., 2008). Increased severity of sarcopenia reduces quality of life since older people cannot accomplish everyday tasks (Rizzoli, 2014). Furthermore, sarcopenia results in a substantial increase in the risk of falls and hip fractures (González-Montalvo et al., 2016) leading to a three-fold increase in mortality (Panula et al., 2011). To facilitate an increase in “active” life-expectancy, lifestyle interventions are paramount, including adopting optimal nutrition strategies combined with increased activity via the use of whole-body exercise (Abate et al., 2017). Esmarck et al., (2001) found that increasing physical activity by engaging in resistance exercise may reduce the risk of falls. The likely mechanisms responsible are the associated increases in muscle hypertrophy (Tipton and Wolfe, 2001) via the stimulation of increased muscle protein synthesis (MPS) (Damas et al., 2015). The muscle mass, and the functional gains observed in younger individuals following resistance training (Moore et al., 2004) appear to be less pronounced in the elderly (Wilkinson et al., 2018). Previous studies have shown that whilst resistance training does have some beneficial effects in the elderly, the anabolic response to exercise is blunted (Malafarina et al., 2013) and has been termed anabolic resistance.

The severity of the outcomes associated with anabolic resistance may be further exacerbated by the lower protein content often observed in the diet of older populations (Evans 2004). Phillips et al., (2016) suggested that older individuals require larger dose supplementation of protein post-exercise compared to younger populations. In a meta-analysis of studies using resistance exercise training and protein supplementation in the elderly, Finger et al., (2015), demonstrated that the combination of these two interventions simultaneously, may have positive effects on fat-free mass but not muscle mass or muscle strength. However, none of the studies included in this meta-analysis used post-exercise protein intakes that were high enough to sufficiently overcome the effects of anabolic resistance. It is also likely that the existing studies have used protein supplementation protocols that do not specifically coincide with exercise stimulated increases in the rate of MPS (Iglay et al., 2009; Leenders et al., 2013) or have used participants that already show signs of reduced mobility (Kim et al., 2012; Chalé et al., 2013). Macnaughton et al., (2016) investigated the provision of either 20 g or 40 g of protein during recovery from a single bout of whole-body resistance exercise in resistance trained males. Their rationale for this ingestion strategy related to the potential need for additional amino acid availability in populations with larger muscle mass, completing whole body exercise (Mcnaughton et al., 2016). They demonstrated that the acute MPS was stimulated to a greater extent when 40 g of whey protein was supplemented via infusion, post-exercise. This threshold is also likely in older individuals, given the likelihood of both symptoms of sarcopenia and anabolic resistance.

At present few studies have considered the effects of post-exercise protein ingestion in the elderly, (Daly et al., 2014; Leenders et al., 2013). This has also not been considered in whole-body exercise training, which is most likely to be recommended in older age groups. Furthermore, none of these studies have used intakes which are high enough to meet the requirements (Moore et al., 2015) nor have they considered this in older individuals that are unaccustomed to resistance exercise. Therefore, the aim of the present study was to determine the effects of a low and high post-exercise dose of whey protein, on anthropometric and functional strength during a 10-week whole body resistance-training programme in elderly males.

Methods

Experimental Approach to the Problem

A randomised group design was used to investigate the effects of resistance training and different whey protein intake, on body composition and strength, in untrained, but recreationally active elderly male participants.

Participants

Twenty-four healthy elderly Caucasian male adults (60-85 years old) volunteered for this study (Fig 1). All participants were recreationally active, undertaking some form of physical activity (walking, golf), 1-2 times per week. None of the participants were undertaking any regular resistance exercise. Volunteers were screened for contraindications to exercise, resulting in 19 participants being enrolled in the study (Fig 1). The exclusion criteria were (1) major surgery in the past 12 months, (2) acute or chronic cardiovascular pathology, (3) history or present use of corticosteroids (including nasal sprays), growth hormone or testosterone, (4) Type 1 or Type II diabetes mellitus, (5) milk protein intolerance or allergies, (6) blood pressure $>$ 140mmHg and/or $>$ 90 mmHg for systolic and diastolic pressure respectively, (7) current use of whey protein supplements. Those participants excluded for being discordant with criteria 1-6 were excluded were referred to their General Practitioner. Participants were then assigned to one of two matched experimental groups, which consumed either 20 g or 40 g of whey protein after resistance exercise training sessions, using a single blind experimental design. Groups were matched based on their age, anthropometric characteristics and habitual protein intake. Prior to data collection, all participants signed written informed consent after being informed of all the procedures and potential risks involved in the investigation. The study was approved by the Departmental Research Ethics Committee (SPA-REC-2015-401).

[Insert Fig 1 near here]

Experimental Procedures

Anthropometric Assessment

Body composition was assessed before the start of the training period and on completion of week 10, using air-displacement plethysmography (Bodpod, Cosmed, Italy), which has previously been reported to be suitable for use in older populations providing a safe, valid and reliable alternative to more traditional body composition techniques (Fields and Hunter, 2004). Calibration of chamber pressure amplitudes occurred before all tests using a 50 L calibration cylinder. Participants attended the laboratory at 09.00 h following a minimum of a 3 h fast and having abstained from exercise in the previous 12 h. Minimal, but tight-fitting clothing and a swim cap, were worn during the procedure in accordance with the manufacturer's guidelines (Bodpod, Cosmed, Italy). Participants were required to follow the same procedures before and during the anthropometric measurement visits.

Determination of Maximal Strength

The first visit to the fitness suite was used to familiarise the participants with the environment, resistance exercise equipment and lifting techniques. This was done by a qualified fitness instructor, who used coaching points in order to ensure correct techniques were used. During this visit participants were also acquainted with the handgrip dynamometer (Takei Scientific Instruments Co., Ltd, Takei 5401 Grip D, Tokyo, Japan) for assessment of grip strength. The subsequent visit was used to make baseline strength measurements. This occurred seven days after the familiarisation session and required participants to attend at 09.00 h having abstained from exercise, alcohol and caffeine for 24 hours (Graham, 2001, Shirreffs and Maughan, 2006). Grip strength was measured using a calibrated hand dynamometer with adjustable grip.

The test was performed in a standing position in accordance with the manufacturer's guidelines (Takei Scientific Instruments Co.). Participants squeezed the dynamometer with as much force as possible for approximately 10 s using their dominant hand. The highest score of three attempts was recorded, with each attempt separated by one minute.

Following a further 5 min rest period, participants then undertook a warm-up by cycling for 4 min against no resistance (TechnoGym Cycle Egometer, Italy). Coaching points for the subsequent lifts were reiterated during the weight based warm up as suggested by the National Strength and Conditioning Association (NSCA) (Earle and Baechle, 2004). Maximal strength, or one repetition maximum (1RM) was then determined using the protocol previously described by Sheridan et al., (2018) which has been widely used to describe maximal strength (Baechle et al., 2008) for seated chest press, leg extension, shoulder press, leg press, and latissimus dorsi (lat) pulldown. This involved the performance of a 12-repetition warm-up set at 10% of estimated 1RM followed by a six-repetition set with an additional 20% load. Then, a pre-maximal set of three repetitions of estimated near maximal load was performed. Participants then attempted single lifts with 1.25, 2.5 or 5 kg progressively added if they were successful in order to determine the final 1RM. Rest periods of 2-4 min were used between lifts. All 1RM's were determined within 3-6 attempts and 5 min rests were used between each 1RM mode of assessment. The order of assessment was repeated on each subsequent visit at week 5 and week 10. Week 5 assessments were used to ensure progressive overload and therefore appropriate progression relative to changes in 1RM.

Resistance Training Programme

The participants completed a supervised progressive resistance-training programme that was performed on non-consecutive days, 3 times a week for 10 weeks (30 training days). A 10-week programme was chosen as increases in skeletal muscle hypertrophy are generally seen after six to eight weeks of resistance training (Staron et al., 1994).

The participants trained collectively in a supervised gym environment on Monday, Wednesday and Friday, ensuring all participants completed all components of each session. Each session commenced with a standardised 10 min warm-up on a cycle ergometer at low resistance followed by a stretching session that was led by a personal trainer. Once completed they performed the resistance-training programme that engaged in whole body exercises as previously described for 1RM assessment. In addition, all participants performed the same resistance training programme which consisted of performing 3 sets of 10 repetitions (reps) for each exercise at 70% of 1RM in accordance with the recommendations for older adults (Fragala et al., 2019). One individual in the 20 g group could not perform the latissimus dorsii pulldown exercise due to a prior injury but was able to undertake all other activities. During all training sessions expert qualified personal trainers supervised each participant.

Supplementation Strategy

Following the initial anthropometric assessments participants were assigned to one of the two matched experimental groups. Habitual protein intake was determined using a 24 h diet diary which was clarified with all participants via telephone, 24 h after they had completed it which has been shown to allow the estimation of protein intake (Beer-Borst and Amadò 1995). The diet diaries were then analysed using dietary analysis software (MicroDiet, Downlea Systems Ltd, UK). Participants then received either a low (20 g) or high (40 g) dose of whey protein (Big Whey, NutritionX, Gloucester, UK – see Table 1 for nutritional information) immediately following each resistance exercise training session. Drinks for every participant were made and administered by gym staff that were not directly part of the research team and were blinded to the nature of the study. Ingestion was supervised to ensure full adherence. This design was chosen to replicate that used by Macnaughton et al., (2016), but this current study focused on training with chronic, rather than the acute ingestion responses. Participants were required to adhere to their normal dietary intake throughout the 10-

week training period, so that the only alteration to dietary intake was the additional whey protein provided after the resistance exercise sessions.

[Insert Table 1 near here]

Statistical Analysis

All data were initially assessed for normality using standard graphical procedures (Grafen and Hails, 2002). Assessment of the differences in 1RM, grip strength and anthropometrical data between the two dose groups, were analysed using analysis of covariance (ANCOVA), with Bonferroni *post hoc* analysis used to determine ingestion strategy main effects. Habitual protein intake comparisons between groups were analysed using an independent t-test. Within group differences across the training period were assessed using paired t-tests. Effect sizes were analysed using partial eta squared (ηp^2) for ANCOVA and Cohen's d_{av} (Lakens, 2013) for t-tests (referred to as d for the remainder of the manuscript). Effect sizes were interpreted as trivial, small, moderate or large using values of <0.20, 0.20-0.49, 0.50-0.79, ≥0.8 respectively (Cohen, 1988). All statistical analyses were conducted using SPSS 24 for Windows, and statistical significance was accepted at the $p < 0.05$ level. All data are reported as mean (±SD).

Results

There were no significant differences in habitual dietary protein intake between the 20 g (57.8 ± 14.5 g.day^{-1} [0.6 ± 0.1 g.kg^{-1}.day^{-1}]) or the 40 g (58.8 ± 13.5 g.day^{-1} [0.6 ± 0.1 g.kg^{-1}.day^{-1}]) group (mean difference [MD] = 1.0 g.kg^{-1}.day^{-1}, t = 0.124, p = 0.904, d = 0.02) prior to the start of supplementation. On completion of the 10-wk training period, one participant in the 20 g group admitted to completing several additional aerobic exercise session each week. This resulted in considerable weight and fat mass losses, we therefore classed this participant as an outlier for his failure to adhere to the prescribed training sessions and removed all of his data from the analysis. This resulted in two n = 9 experimental groups (Fig 1). There were no significant differences between group effects on any of the anthropometric variables (Table 2) in responses to the different protein ingestion doses ($F = 0.001$, $p = 0.981$, ηp^2 0.00, $F = 0.496$, $p = 0.492$, ηp^2 0.03, $F = 0.557$, $p = 0.467$, ηp^2 0.04, $F = 0.955$, $p = 0.344$, ηp^2 0.06, for body mass, FFM, FM and body fat respectively). However, there were some within group changes to the anthropometric variables in response to the training programme. In the 20 g group FM (MD = 1.73 kg, $t = 4.40$, $p = 0.002$, $d = 0.17$) and BF (MD = 1.34%, $t = 6.32$, $p < 0.001$, $d = 0.21$) significantly decreased, and in the 40 g group, body mass (MD = 1.46 kg, $t = 2.84$, $p = 0.022$, $d = 0.09$), FM (MD = 1.28 kg, $t = 3.66$, $p = 0.006$, $d = 0.12$), and BF (MD = 0.93%, $t = 2.75$, $p = 0.025$, $d = 0.12$).

[Insert Table 2 near here]

Interestingly, 1RM performance was increased to a significantly greater amount in the 40 g group for chest press ($F = 7.72$, $p = 0.014$, ηp^2 0.34; Fig 2a), shoulder press ($F = 11.10$, $p = 0.005$, ηp^2 0.43; Fig 2b) and leg extension ($F = 7.66$, $p = 0.014$, ηp^2 0.34; Fig 2c) compared with the 20 g group; but this was not the case for lat pulldown ($F = 0.27$, $p = 0.612$, ηp^2 0.02; Fig 3a), leg press ($F = 2.27$, $p = 0.153$, ηp^2 0.13; Fig 3b) or grip strength ($F = 1.08$, $p = 0.315$, ηp^2 0.07; Fig 3c), (. This observation occurred even though there were no significant pre-intervention differences in any of the 1RM assessments (Fig 2 and Fig 3).Significant post training improvements were observed for chest press (MD = 6.33 kg, $t = 5.49$, $p = 0.001$, $d = 0.30$ and MD = 16.56 kg, $t = 4.84$, $p = 0.001$, $d = 0.67$), shoulder press (MD = 4.89 kg, $t = 3.90$, $p = 0.005$, $d = 0.25$ and MD = 11.78 kg, $t = 4.65$, $p = 0.002$, $d = 0.46$) and leg extension (MD = 10.63 kg, $t = 3.98$, $p = 0.005$, $d = 0.18$ and MD = 13.44 kg, $t = 3.67$, $p = 0.006$, $d = 0.87$) lat pulldown (MD = 0.93 kg, $t = 3.58$, $p = 0.007$, $d = 0.39$ and MD = 0.93 kg, $t = 3.67$, $p = 0.006$, $d = 0.74$), leg press (MD = 0.93 kg, $t = 4.59$, $p = 0.002$, $d = 0.28$ and MD = 0.93 kg, $t = 4.61$, $p = 0.002$, $d = 0.37$) and grip strength (MD = 0.93 kg, $t = 2.52$, $p = 0.036$, $d = 0.12$ and MD = 0.93 kg, $t = 3.81$, $p = 0.005$, $d = 0.22$), in both the 20 g and 40 g protein groups respectively. There were also significant differences in the percentage improvements in 1RM for chest press, shoulder press and leg extension (Table 3).

[Insert Figure 2 and 3 near here]

[Insert Table 3 near here]

Discussion

The purpose of this study was to investigate the combined effects of resistance training with either low or high dose post-exercise protein ingestion, in older males, novice to exercise training. The study design allowed careful and close supervision of participants in all sessions, to ensure both adherence to the training programme, and ingestion of the supplements immediately after each training bout. The key findings suggest that the provision of 40 g of whey protein immediately after exercise provides an additional functional training response to that of a 20 g dose in some muscle strength assessments, following a ten-week resistance training programme in older adults. These findings are in contrast to those of a recent systematic review (Thomas et al., 2016), which suggested that additional supplementation with protein did not augment the effects of resistance training in older participants. Furthermore, a meta-analysis by Finger et al., (2015) suggested that protein ingestion when combined with resistance training was only effective for eliciting improvements to FFM, but not muscle mass nor strength. Conversely, Liao et al., (2017) demonstrated that protein supplementation and resistance training may provide a stronger effect than resistance training alone on reducing the attenuated muscle mass loss, and leg strength. The seemingly paradoxical findings of these reviews and meta-analyses are likely explained by the relatively low doses of protein ingested, the timing of ingestion not always coinciding with the end of exercise and the wide variety of protein sources ingested in the studies included in these analyses. Indeed, Mcnaughton et al., (2016) observed more pronounced rates of MPS when 40 g of protein was observed acutely, suggesting that this higher dose may be more optimal (Witard et al., 2016).

The findings of significantly improved strength performance in chest, shoulder and leg press, can be accounted for by the use of immediate post-exercise ingestion of the protein supplements and the relatively large dose in the 40 g group, since there was no difference in the pre-intervention protein intakes between the groups. These findings add further support to the suggestion that the negative impact of age-related protein resistance can be off-set by simply providing a higher dose of protein (Murphy et al., 2015) to older individuals, ideally immediately following resistance exercise (Macnaughton et al., 2016; Witard et al., 2016). Interestingly, there were no observed significant changes to grip strength, which might have been expected. In a recent meta-analysis Labott et al., (2019) calculated a likely small effect of training in older adults (SMD = 0.28 [95% CI = 0.13, 0.44]). These authors also suggest that a meaningful change in grip strength performance is likely ~1.6 kg, but possibly up to 3.6 kg. Given that none of the participants in the present study achieved post-training improvements higher than 2.6 kg and our calculated co-efficient of variation for this assessment is was 3.4%, it was unlikely these would be meaningfully different. Indeed, just five participants performed better than the test variability.

The strength improvement observed in both groups are the likely result of neuromuscular adaptations which occur in response to repeated training (Close et al., 2008). Rapid increases in strength have been observed during the first few weeks of training in the elderly (Mayer et al., 2011), due to neural adaptation mechanisms associated with increased force production (Reeves et al., 2003) at magnitudes of improvement similar to those in the 20 g group. Indeed, improvement in 1RM performance of ~19% are typically reported for training programmes of similar lengths in the elderly (Reeves et al., 2003; Hakkinen et al., 1996), suggesting that the low dose of protein had limited effects on the 1RM performance in the present study.

The responses observed in the 40 g group could be due to the combined effects of exercise and protein, especially leucine, creating a more optimal stimulation of muscle protein synthesis (Traylor et al., 2018), but this occurred independently of significant increases in mass, suggesting that a longer intervention, may be more beneficial. Previously Moore et al., (2015), suggested that 0.4 $g.kg^{-1}$ was an optimal protein dose per meal in older adults and it is this in part that has informed the call for minimum daily intake recommendations for protein in older adults to be increased to 1.2 $g.kg^{-1}.day^{-1}$ (Traylor et al., 2018). In the present study the doses equated to 0.22 ± 0.03 and 0.46 ± 0.09 $g.kg^{-1}$ for the 20 g and 40 g doses respectively. Importantly, the 40 g dose is over the minimal dose threshold of 0.4-0.6 $g.kg^{-1}$ which is needed to maximally stimulate MPS when protein is ingested as part of a mixed meal (Kim et al., 2012). Crucially, the co-ingestion of mixed macronutrient meals has been shown to increase the amount of protein needed to maximally stimulate MPS, due to changes in amino acid absorption and uptake (West et al., 2011). Therefore, provision of solely a protein supplement in the present study, is likely to be over the minimal amount required to maximally stimulate MPS. We do however acknowledge that this is a research informed assumption, since we were unable to directly measure the rate of MPS. However, it is also likely that the relatively low normal intake of protein in both groups was improved with ingestion strategy, and resulted in the mean protein intakes being more optimal (0.8 ± 0.1 $g.kg^{-1}.day^{-1}$ and 1.0 ± 0.1 $g.kg^{-1}.day^{-1}$ for the 20 g and 40 g groups respectively) on exercise days.

The other limitation to the present study is the length of the resistance training intervention, which may not have been long enough for the observed strength improvements and training adaptations, to be reflected in FFM increases. However, within-group body fat and fat mass were significantly reduced in both groups, with a greater weight loss in the 40 g group, which warrant further investigation. The magnitude of change in these body compositional variables is commensurate with those previously reported (Liao et al., 2017), and may have been limited by the sensitivity of the measurements available and by initial body composition of the participants at the start of the present study. Future work should therefore consider using longer interventions to determine the possible impact of post resistance training, high dose protein ingestion on changes to muscle mass in older adults. Furthermore, in any future work, quality of life and daily living tests should be included to help determine whether these are improved by using such a nutritional strategy. However, this simple nutritional strategy could have considerable importance for the augmentation of adaptations to training in elderly populations who are prescribed resistance exercise.

Conclusion

The key findings of the present study show that supplying 40 g of whey-based protein immediately following resistance exercise sessions to elderly Caucasian males, is likely to augment beneficial strength and body compositional adaptations via reductions in fat mass. These adaptations have the potential to have a considerable impact on risk factors associated with poor quality of life and disease-free living duration, but more work is needed to establish if the same beneficial responses can be observed in females, and if the effects of this nutritional intervention are amplified during longer term training programmes.

Practical Applications.

Elderly individuals participating in recreational activity, should be aware that consuming 40 g of whey immediately after activity, is potentially more beneficial than 20 g at improving strength and functional activity. This should help such individuals have an improved quality of life.

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Availability of Data and Materials

The datasets used and/or analysed during the current study are available from the corresponding author on reasonable request.

Authors' contributions

CA, GLC and SAS conceived the study. CA recruited participants and collected the data. SAS performed the statistical analysis. CA, LRM, GLC and SAS contributed to drafts of the manuscript, and all authors have read and approved the final version of the manuscript.

Declaration of Interests

CA, LRM and SAS declared no potential conflicts of interest with respect to the research, authorship and/or publication of this article. GLC has previously worked for Nutrition X.

Consent for Publication and Ethical Approval

The authors declare that they consent to publication for the manuscript in the present form. This study was approved by the Department of Sport and Physical Activity Research Ethics Committee, Edge Hill University. All participants provided written informed consent to take part in the study.

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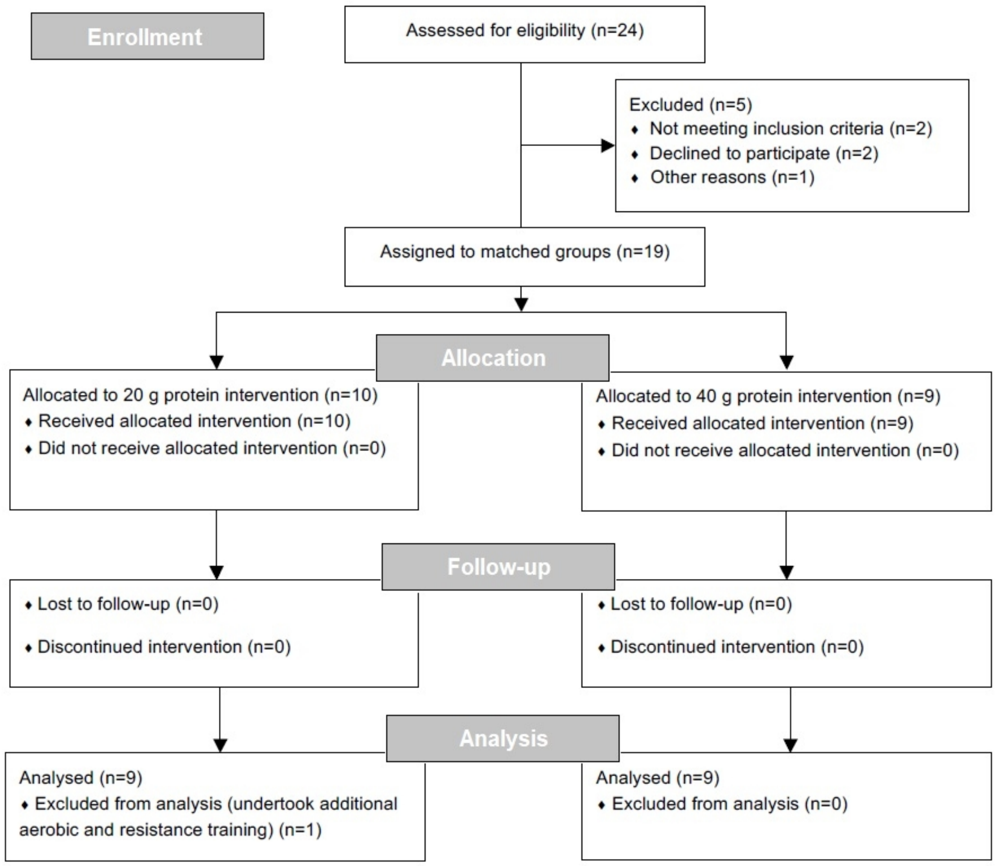

Figure 1 CONSORT flowchart of participant recruitment, experimental group allocation and study design.

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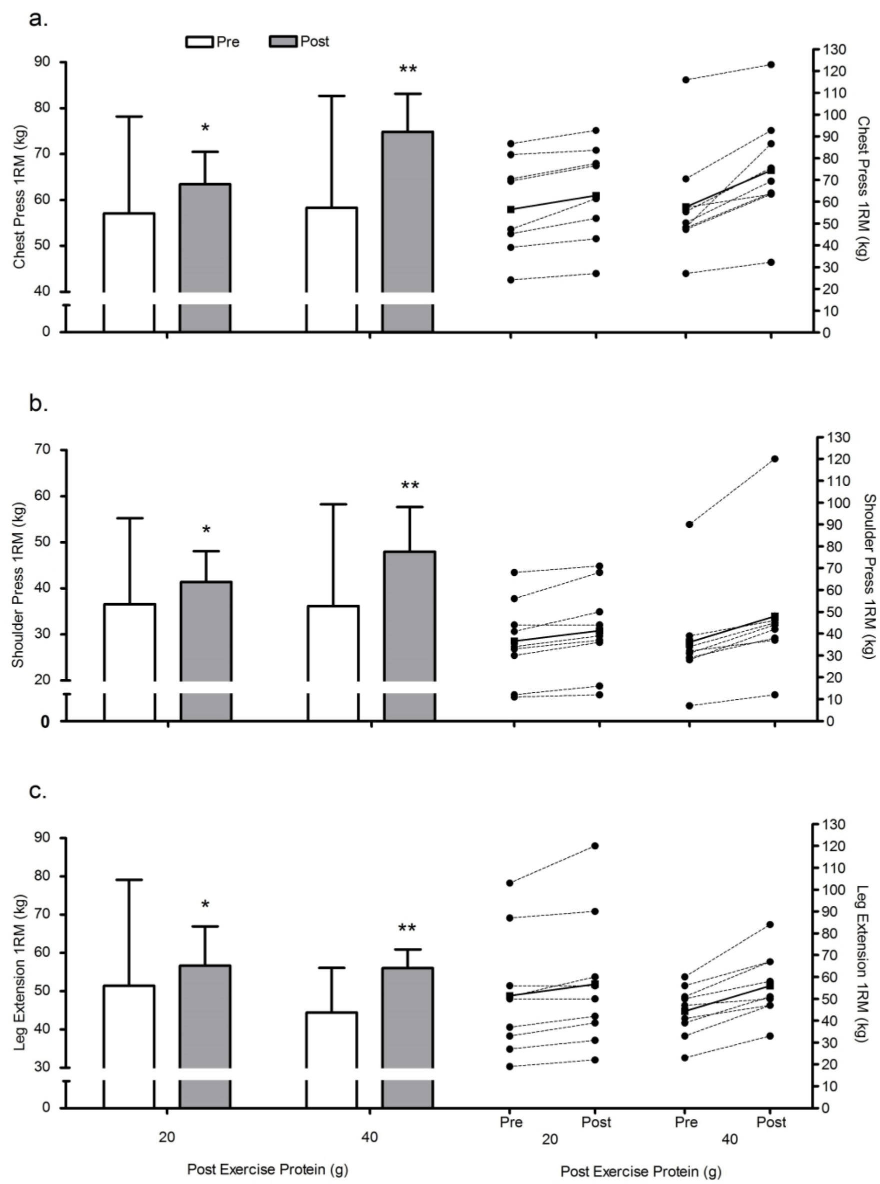

Figure 2 Mean (±SD) and individual chest press (a), shoulder press (b), and leg extension (c) 1RM responses to 10 weeks of resistance training in both protein dose groups. (*) denotes a significant increase in 1RM after training, p<0.05. (**) denotes a significantly greater increase in 1RM in the 40 g condition and a significant increase after training (p < 0.05).

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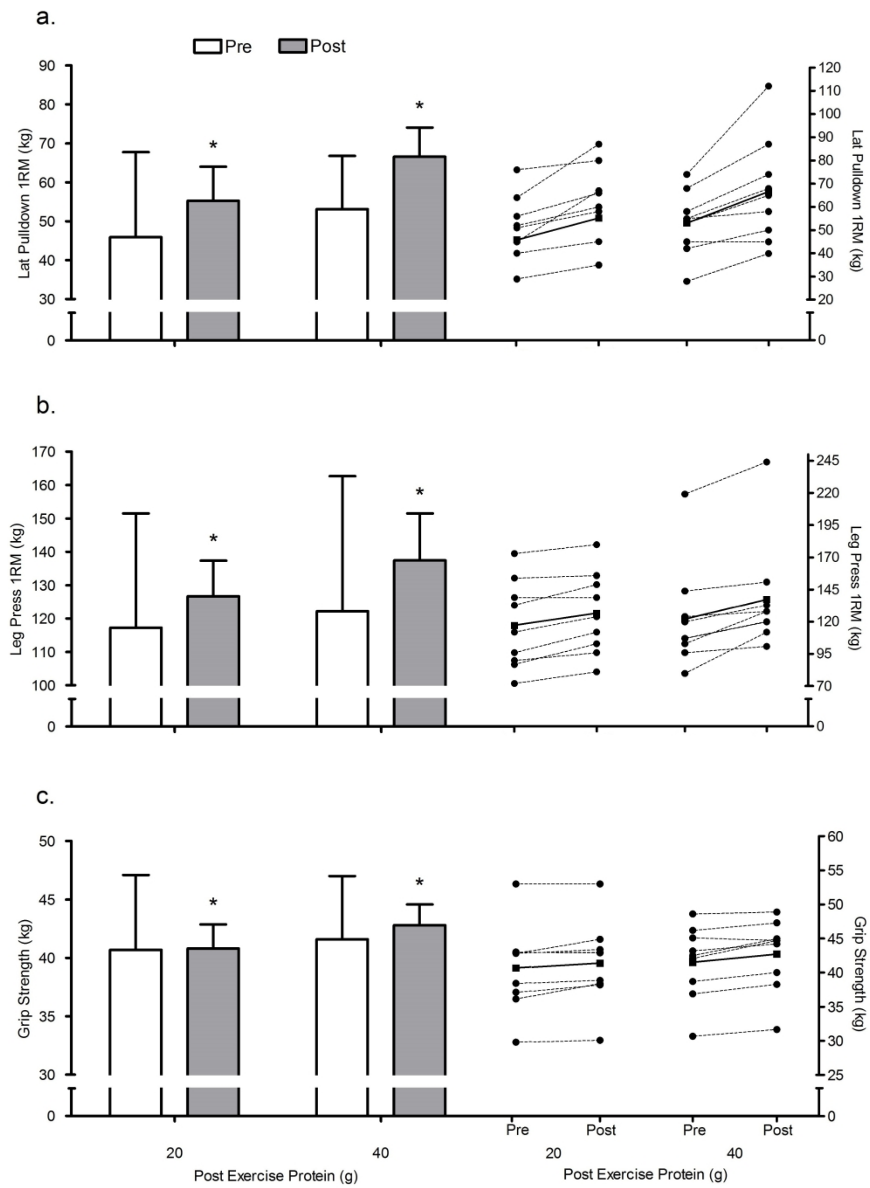

Figure 3 Mean (±SD) and individual lat pulldown (a), leg press (b), and grip strength (c) 1RM responses to 10 weeks of resistance training in both protein dose groups. (*) denotes a significant increase in 1RM after training, $p<0.05$.

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Table 1. Nutritional information for the two post-exercise protein doses.

	Protein Portion (g)	
Nutrient	**20**	**40**
Energy		
kJ	408.1	816.2
kcal	96.5	192.9
Fat	1.4	2.8
Carbohydrate	2.0	3.9
of which sugars	1.2	2.3
BCAA's	5.3	10.6

Table 2. Anthropometric responses to the 10-week training programme in the 20 g and 40 g groups and typical error of measurements (TEM). (*) denotes a significant within group difference between week 1 and 10 ($p < 0.05$). Diff (%) represents the mean percentage difference within groups. ES represents the Cohen's d effect size.

	Experimental Group								
	20 g				40 g				
Variable	Pre	Post	Diff (%)	ES	Pre	Post	Diff (%)	ES	TEM (%)
Mass (kg)	94.7 ± 14.5	93.3 ± 14.8	-1.6 ± 2.3	0.10	90.1 ± 16.6	88.6 ± 16.3*	-1.6 ± 1.7	0.10	0.15
FFM (kg)	62.4 ± 7.7	62.7 ± 8.1	0.4 ± 1.8	0.02	60.5 ± 8.4	60.3 ± 8.4	-0.3 ± 1.9	0.02	3.65
FM (kg)	32.6 ± 10.2	30.8 ± 10.0*	-5.6 ± 4.1	0.17	29.8 ± 11.2	28.5 ± 11.1*	-4.6 ± 3.8	0.12	7.58
Body fat (%)	33.8 ± 6.3	32.5 ± 6.4*	-4.1 ± 2.3	0.21	32.3 ± 7.6	31.4 ± 7.6*	3.0 ± 3.1	0.12	6.95

Table 3. Mean (±SD) percentage functional differences following 10 weeks of post resistance exercise protein ingestion. Where difference represents the percentage difference between the groups; (t) is the independent t-test statistic; (p) is the probability value; (ES) is the Cohen's d effect size; (*) a significant difference between groups where $p < 0.05$.

	Increase in performance (%)			Analysis Outcomes		
1RM	20 g	40 g	Difference	t	p	ES
Chest Press	12.3 ± 7.4	31.4 ± 20.8	19.1 ± 21.0*	2.59	0.020	1.35
Shoulder Press	15.2 ± 10.2	36.3 ± 16.9	21.1 ± 18.9*	3.21	0.005	1.56
Lat Pulldown	18.9 ± 15.1	24.5 ± 16.1	5.6 ± 14.1	0.76	0.459	0.36
Leg Press	9.1 ± 6.5	13.8 ± 11.6	4.7 ± 13.8	1.04	0.312	0.51
Leg Extension	11.1 ± 7.7	27.2 ± 13.5	16.1 ± 17.3*	3.11	0.007	1.52
Grip Strength	2.0 ± 2.3	3.0 ± 2.3	1.0 ± 2.7	0.94	0.360	0.44